Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Cancer research, Merck Serono, Karyopharm Therapeutics. Advisory Board; Celgene. RTOG. A.J. Walker: Consultant; AstraZeneca.

2470

Evaluating the Immunomodulatory Profile and Treatment Efficacy of Whole Lung Low-Dose Radiation Therapy (LDRT) in a Preclinical Model of Severe Viral Pneumonia

N. Dhanota,[1] K.A. Pilones,[1] J. Kraynak,[1] S. Demaria,[2] S.C. Formenti,[2] M. Salvatore,[1] and A.E. Marciscano[1]; [1]*Weill Cornell Medical College, New York, NY,* [2]*NewYork-Presbyterian/Weill Cornell Medical Center, New York, NY*

Purpose/Objective(s): Whole Lung Low-Dose Radiation Therapy (LDRT) is currently being tested as a treatment for SARS-CoV-2. Whether LDRT alters the natural history of viral infection, lung microenvironment or host immunity is currently unknown. We developed a preclinical model of severe viral pneumonia using pandemic-strain influenza A virus (IAV) to evaluate potential immunomodulation and treatment efficacy of LDRT.

Materials/Methods: Female BALB/c mice were infected with an intranasal challenge of 2×10^2–2×10^4 PFU of 2009/H1N1 IAV. LDRT was administered at 0.5 Gy or 1.5 Gy x 1 (or sham) 72 hours after IAV challenge. Mice were serially monitored for survival and morbidity (raw lung weight, % weight loss, disease severity index). Acute changes in the lung microenvironment were assayed 72hrs post-LDRT (day 6) with profiling of myeloid/T-cell subsets by cytometry, gene expression by qRT-PCR and histological evaluation of H&E-stained lung sections.

Results: LDRT did not extend survival in lethally challenged (2×10^4 PFU) mice relative to non-irradiated controls [median survival(days): 7 IAV/sham vs 8 IAV/0.5 Gy vs 8 IAV/1.5 Gy; $P = 0.24$], with 100% mortality in all IAV groups. In a less virulent model (2×10^3 PFU), LDRT did not extend survival with significantly shorter survival observed in the IAV/1.5 Gy group [median survival (% mortality): NR (30%) IAV/sham vs 10d (53%) IAV/0.5 Gy vs 9d (87%) IAV/1.5 Gy; $P < 0.001$]. Raw lung weight was ~2-fold higher in IAV mice [mean weight: 0.17g PBS/sham vs 0.31g IAV/sham vs 0.30g IAV/0.5 Gy vs 0.31g IAV/1.5 Gy; $P < 0.01$ all IAV groups]. Percentage change in body weight was -20.8% (IAV/sham), -21.7% (IAV/0.5 Gy) and -22.5% (IAV/1.5 Gy) compared to PBS-control mice on day 6. Immune profiling demonstrated monocytic and neutrophilic lung infiltrate in response to IAV, with significant increases in CD11b+Ly6G+ neutrophils ($P = 0.02$) and Ly6C+CD11b+ classical monocytes ($P = 0.02$). Relative to PBS-challenge, bulk RNA analysis demonstrated robust interferon expression (**Ifnb1, Ifng**) and upregulation of myeloid/T-cell chemotaxis (**Ccl2, Cxcl10**) [$P < 0.05$ in all IAV groups]. A mixed inflammatory response was noted with significant increase in pro- and anti-inflammatory cytokines (**Il6, Il10**) and M1 markers (**Inos2**, **Cd80**) [$P < 0.05$ in all IAV groups]. **Arg1** expression was increased in IAV mice treated with 1.5 Gy LDRT relative to IAV/0.5 Gy ($P = 0.02$) and IAV/sham ($P = 0.02$). Histological evidence of alveolar septum rupture, peri-bronchial infiltration, lung parenchyma destruction and vascular congestion was consistent with severe acute lung injury; similar changes were observed in LDRT and non-irradiated lungs of IAV mice.

Conclusion: In this preclinical IAV model of severe viral pneumonia we did not observe a therapeutic effect of LDRT on survival and morbidity. LDRT did not appear to consistently reduce or reverse IAV-induced inflammatory changes in the lung microenvironment.

Author Disclosure: N. Dhanota: None. K.A. Pilones: None. J. Kraynak: None. S. Demaria: Research Grant; Lytix Biopharma. Consultant; Lytix Biopharma, Ono Pharmaceuticals. S. Formenti: Research Grant; Bristol Myer Squibb, Merck, Varian. Advisory Board; Astra Zeneca, Bayer, Bristol Myers Squibb, Eisai, Elekta, EMD Serono/Merck KGa, GlaxoSmithKline, Janssen, MedImmune, Merck US, Regeneron, Varian, ViewRay. M. Salvatore: None. A.E. Marciscano: None.

2471

Mechanism of Increased Treg Frequency Induced by Irradiated Esophageal Squamous Cell Carcinoma

Y. Wang,[1] T. Li,[2] J. Lv,[2] and L. Xiao[1]; [1]*School of Medicine, University of Electronic Science and Technology of China, Chengdu, China,* [2]*Department of Radiation Oncology, Sichuan Cancer Hospital& Institution, Sichuan Cancer Center, School of Medicine, University of Electronic Science and Technology of China, Chengdu, China*

Purpose/Objective(s): The infiltration of CD4+CD25+Foxp3+ regulatory T cells (Treg) in the tumor microenvironment is one of the main reasons for radiation resistance and tumor recurrence after radiotherapy. It has been established that Treg is more resistant to radiation than other T cells, but the proliferation of immune cells after radiotherapy is affected by other factors, including tumor cells. Treg frequency in the tumor microenvironment after radiotherapy has not been defined. We studied the effect and mechanism of increased Treg frequency induced by irradiated ESCC cell, TE-1.

Materials/Methods: After 2 Gy irradiation, TE-1 cells were co-cultured with normal peripheral blood lymphocytes for 48 hours. Flow cytometry was used to detect Treg/CD4+T cell frequency. The mRNA expression of TGF-β1 and TGF-β2 in TE-1 was detected by qPCR, and the protein content of TGF-β1 and TGF-β2 in the medium was detected by ELISA.

Results: Compared with non-irradiation group, the expression of TGF-β1 and TGF-β2 in TE-1 cells of irradiation group increased, and the protein content of TGF-β1 and TGF-β2 in culture medium increased, the difference was statistically significant ($P < 0.001$). Flow cytometry showed that CD4+CD25+/CD4+Tcell and CD4+CD25+Foxp3+/CD4+Tcell were increased in the radiotherapy group after co-culture, and the difference was statistically significant ($P < 0.001$).

Conclusion: The expression of TGF-β1 and TGF-β2 in esophageal squamous cell carcinoma cells increased after irradiation, and the frequency of Treg induced by co-culture increased, suggesting that esophageal squamous cell carcinoma cells after radiotherapy can induce the increase of Treg cells, which may be achieved mainly through the mechanism of increasing the secretion of TGF-β1 and TGF-β2.

Author Disclosure: Y. Wang: None. T. Li: None. J. Lv: None. L. Xiao: None.

2472

AMG-510 Plus Cetuximab Enhance Radiosensitivity in KRAS p.G12C Mutant Colorectal Carcinoma Cell Lines via Increasing Apoptosis and Inducing G1/S Arrest

T. Lv,[1] Y. Wang,[2] and Z. Zhang[2]; [1]*Department of Radiation Oncology, Fudan University, Shanghai Cancer Center, Shanghai, China,* [2]*Department of Radiation Oncology, Fudan University Shanghai Cancer Center, Shanghai, China*

Purpose/Objective(s): KRAS p.G12C mutation is rare (~3%) but with a dismal prognosis in colorectal carcinoma (CRC). AMG-510 is a first-in-class KRAS p.G12C inhibitor, which shows clinical efficacy in KRAS p.G12C mutant solid tumors including non-small cell lung cancer and CRC. The addition of cetuximab could revert resistance to AMG-510 in CRC. Herein, we evaluate the anti-cancer effect of AMG-510 plus cetuximab in combination with radiation in CRC.

Materials/Methods: Two KRAS p.G12C mutant CRC cell lines were treated with AMG-510, AMG-510 plus cetuximab, irradiation (IR), and the combination of IR and AMG-510 plus cetuximab. Clonogenic assays were used to study the radiosensitizing effect of AMG-510 combined with cetuximab. Immunofluorescence staining of rH2AX was used to detect double-strand break (DSB) repair. Cell proliferation was performed using a cell counting kit. Then, we performed flow cytometry analysis to detect cell apoptosis rate and cell cycle distribution.